

Fig.1.

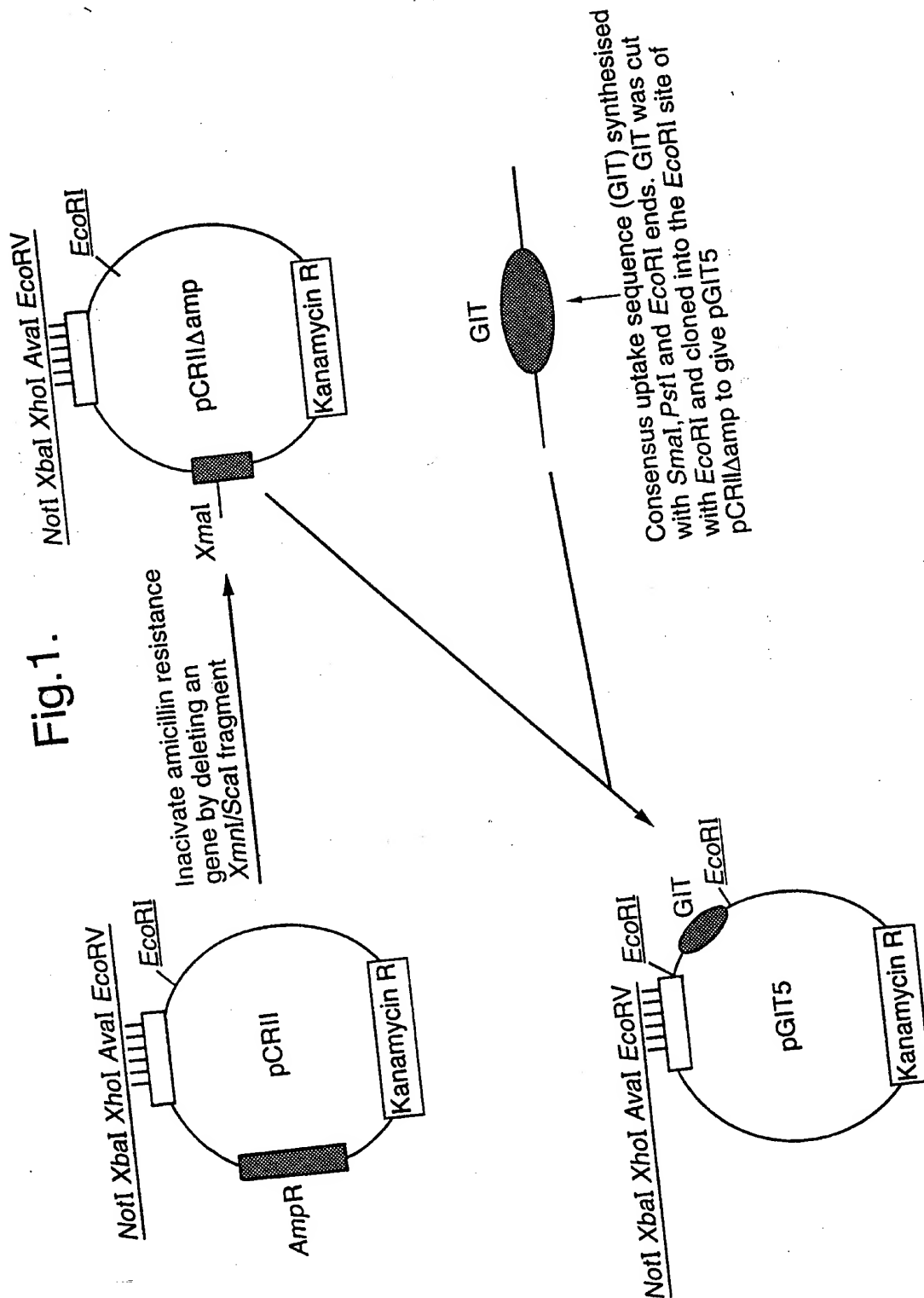


Fig.2.

aroB1-	gcagatgcccgaagcttttatagcgg HindIII	melting temperature=74°C
aroB2-	gagctgggtaccgtgcagcgtgtccagatctgcaag SacI KpnI BglII	melting temperature=72°C
aroB3-	cataaagggtaccgtgttcgccagc BamHI	melting temperature=70°C
aroB4-	ggtaccgagctccaaatgaaggcagatctctgcgcc KpnI SacI BglII	melting temperature=74°C

Amplify the two halves of the *aroB* region by PCR using the above primers in the following combinations

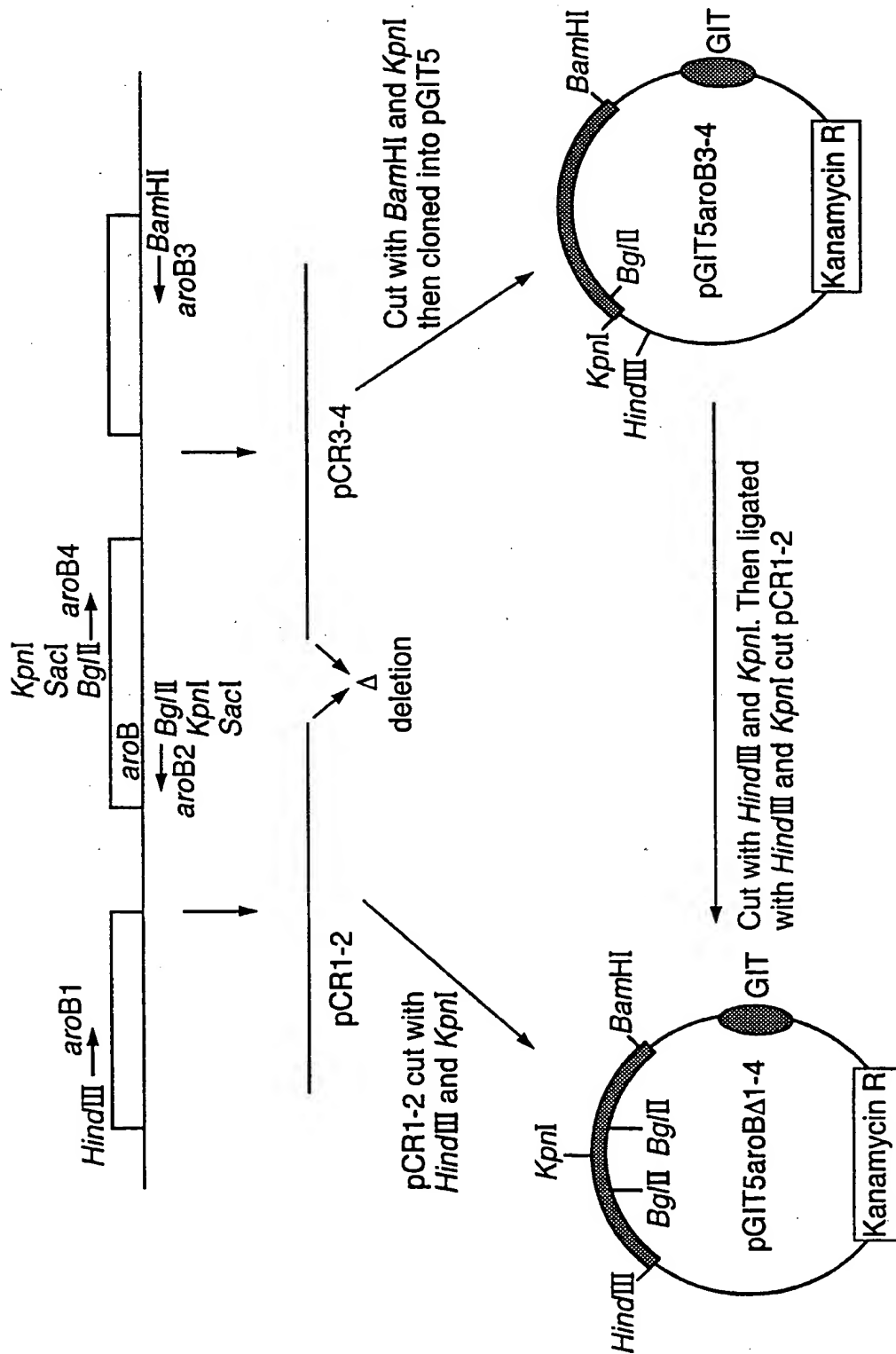
aroB1 + *aroB2* - gives a fragment of 1575 bp. From upstream of *aroB*.

aroB4 + *aroB3* - gives a fragment of 1433 bp. From downstream of *aroB*.

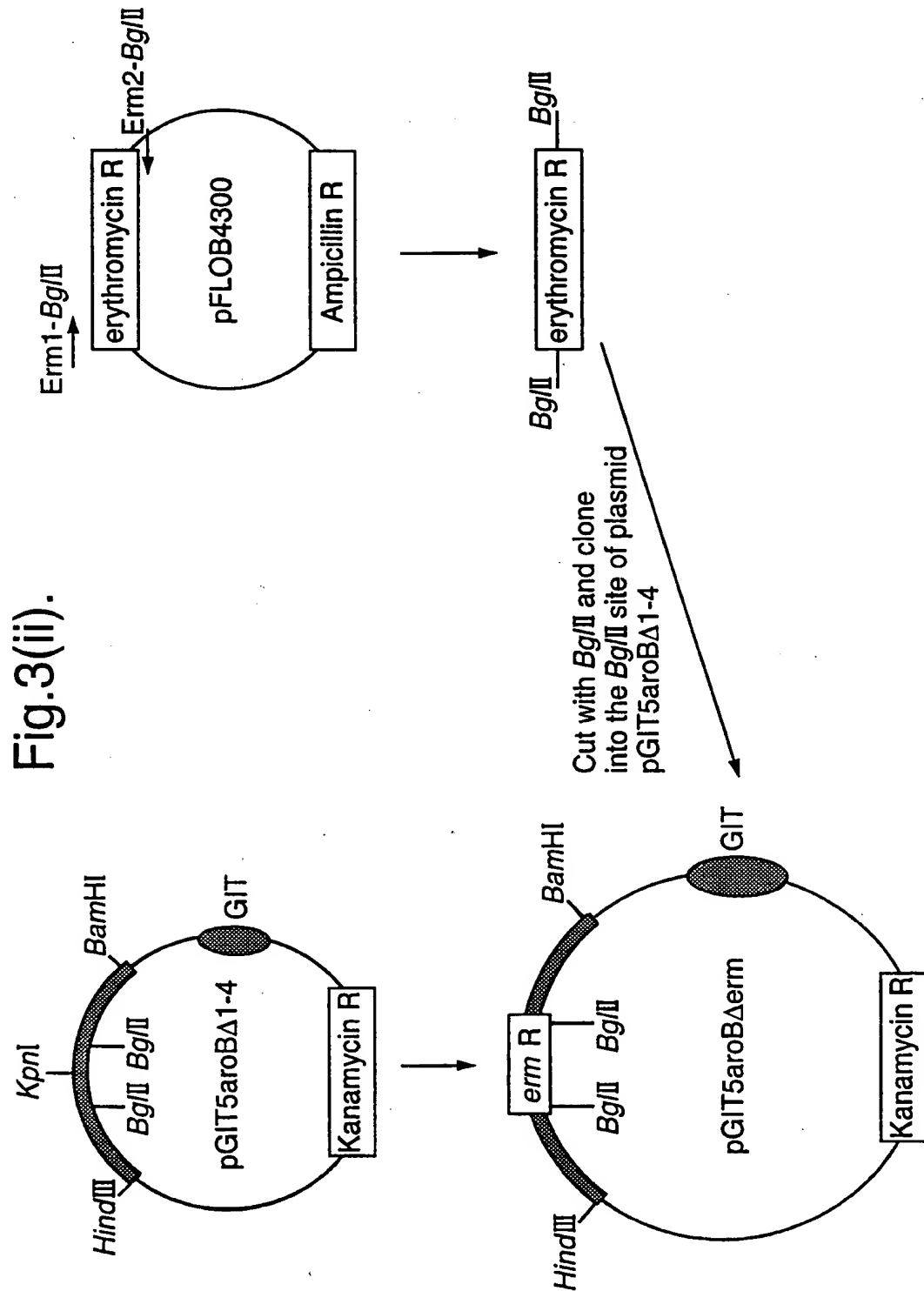
Ligation of these fragments together gives a deletion in the middle of *aroB* of approximately 150 bp.

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Fig.3(i).

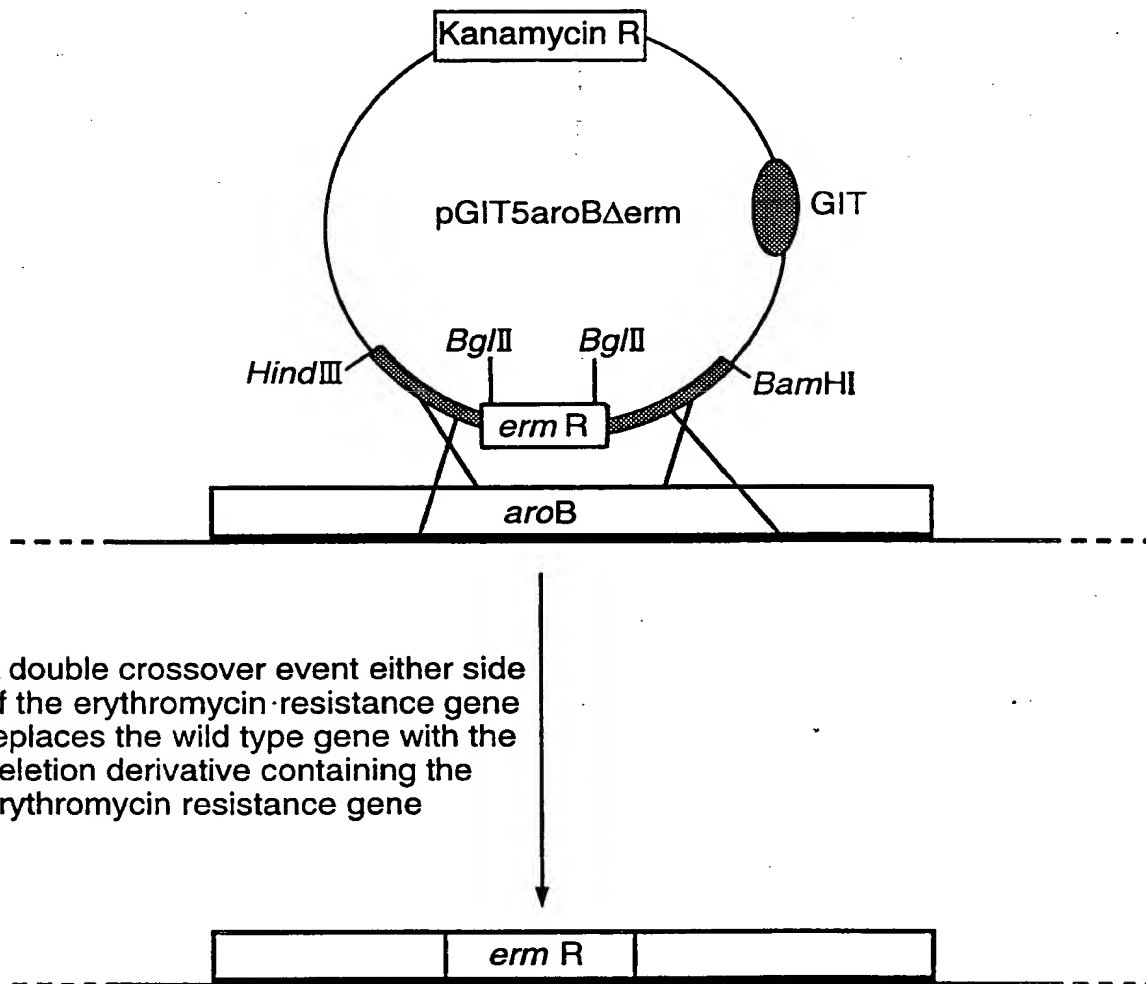


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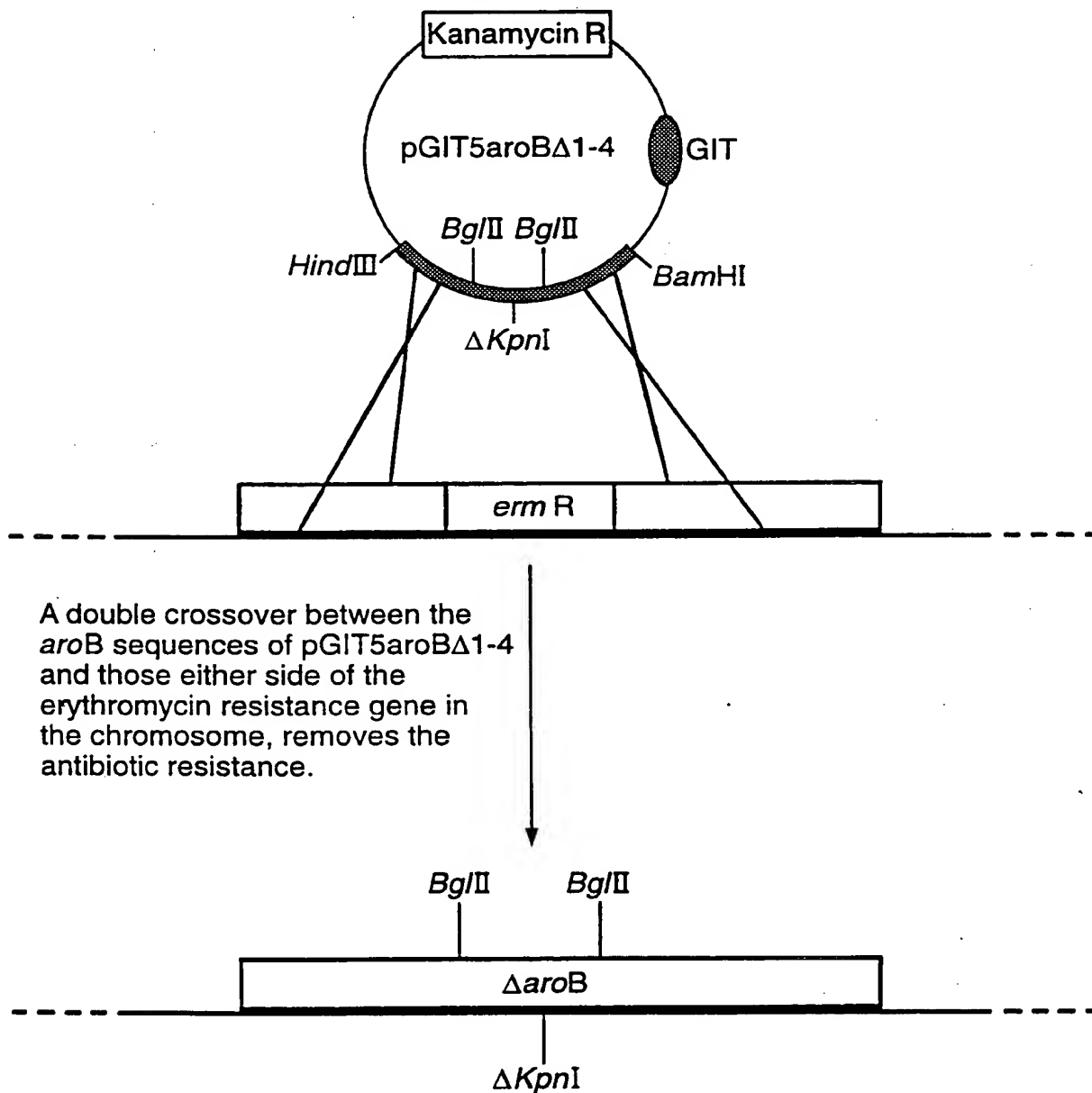
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Fig.3(iii).



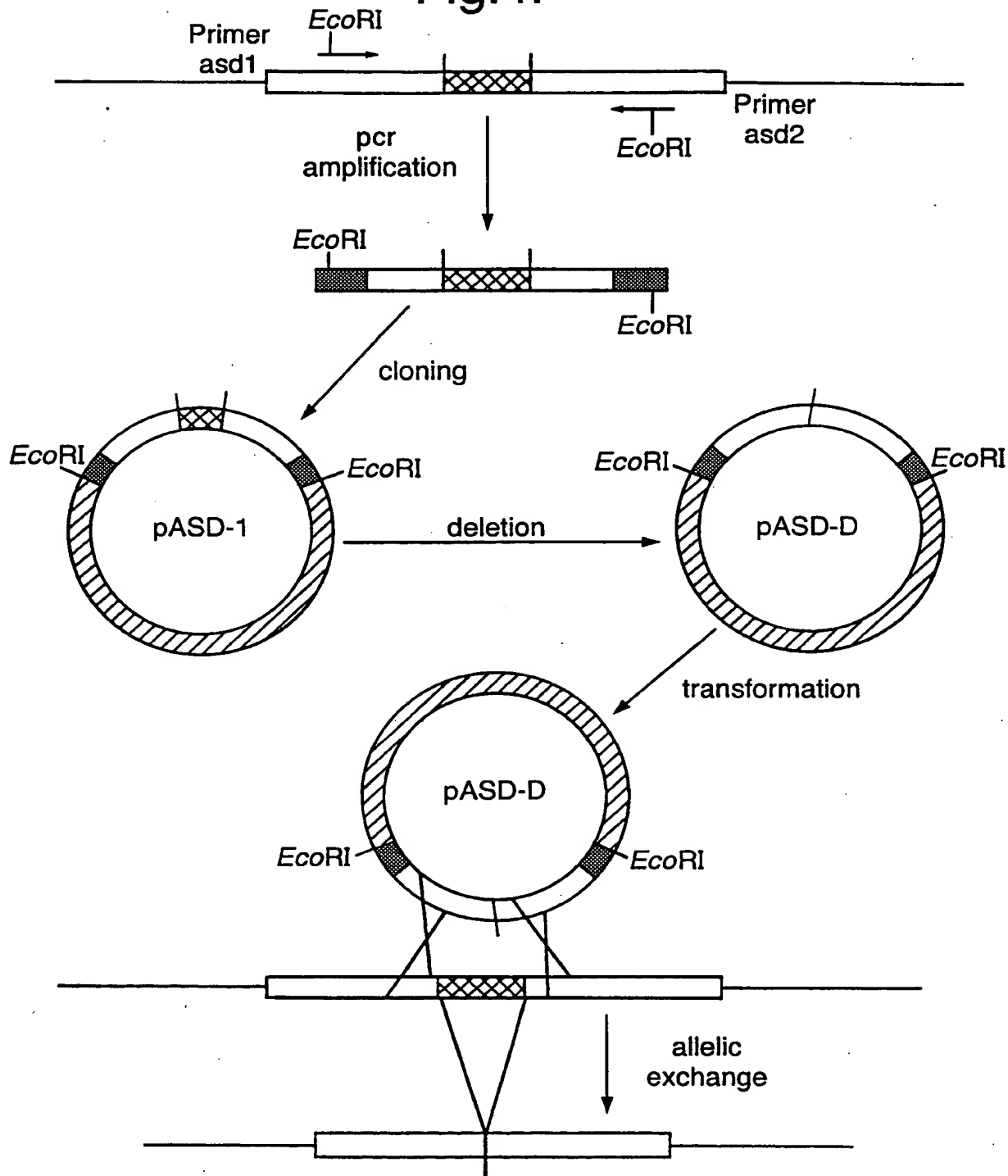
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Fig.3(iv).



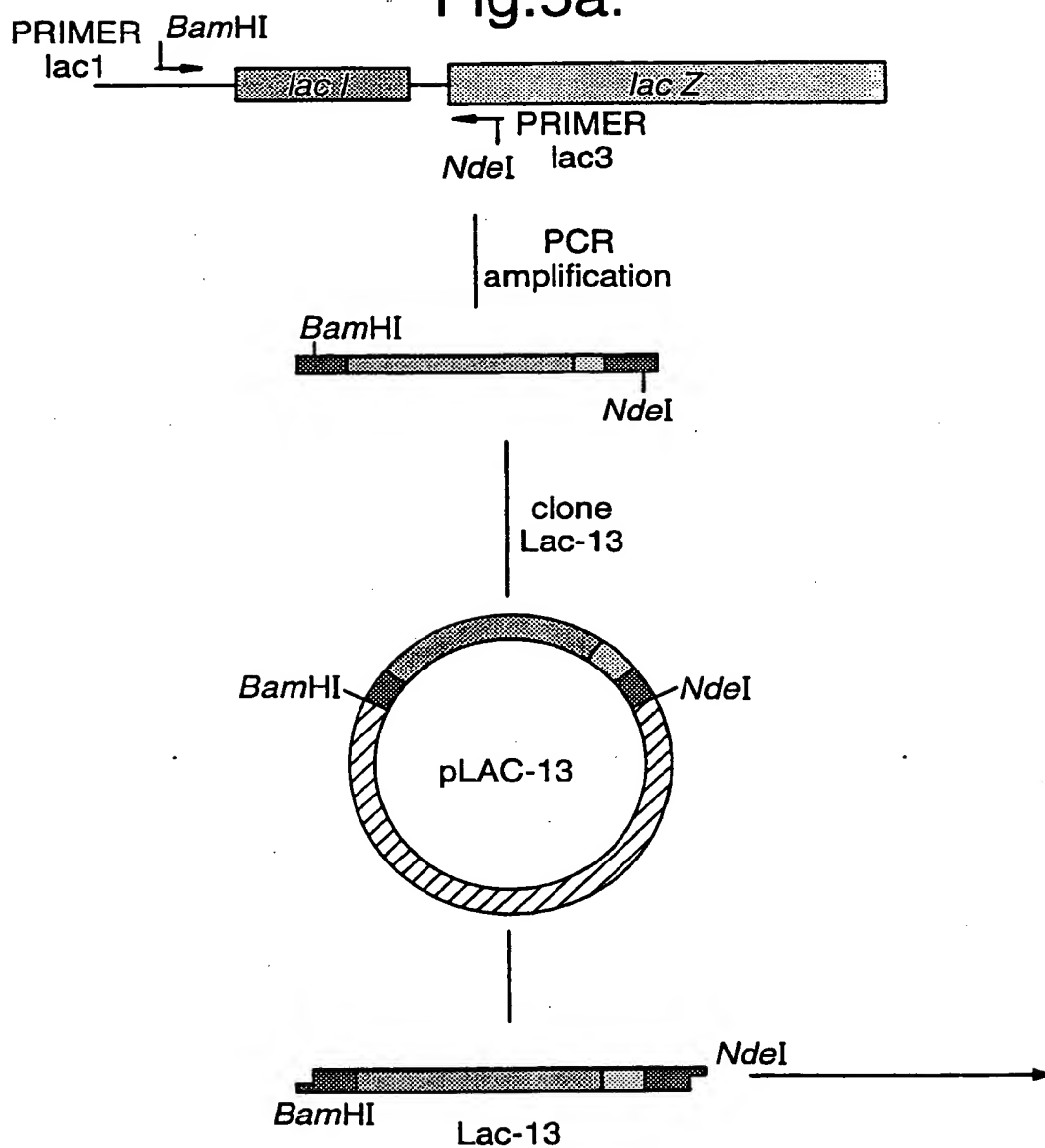
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Fig.4.



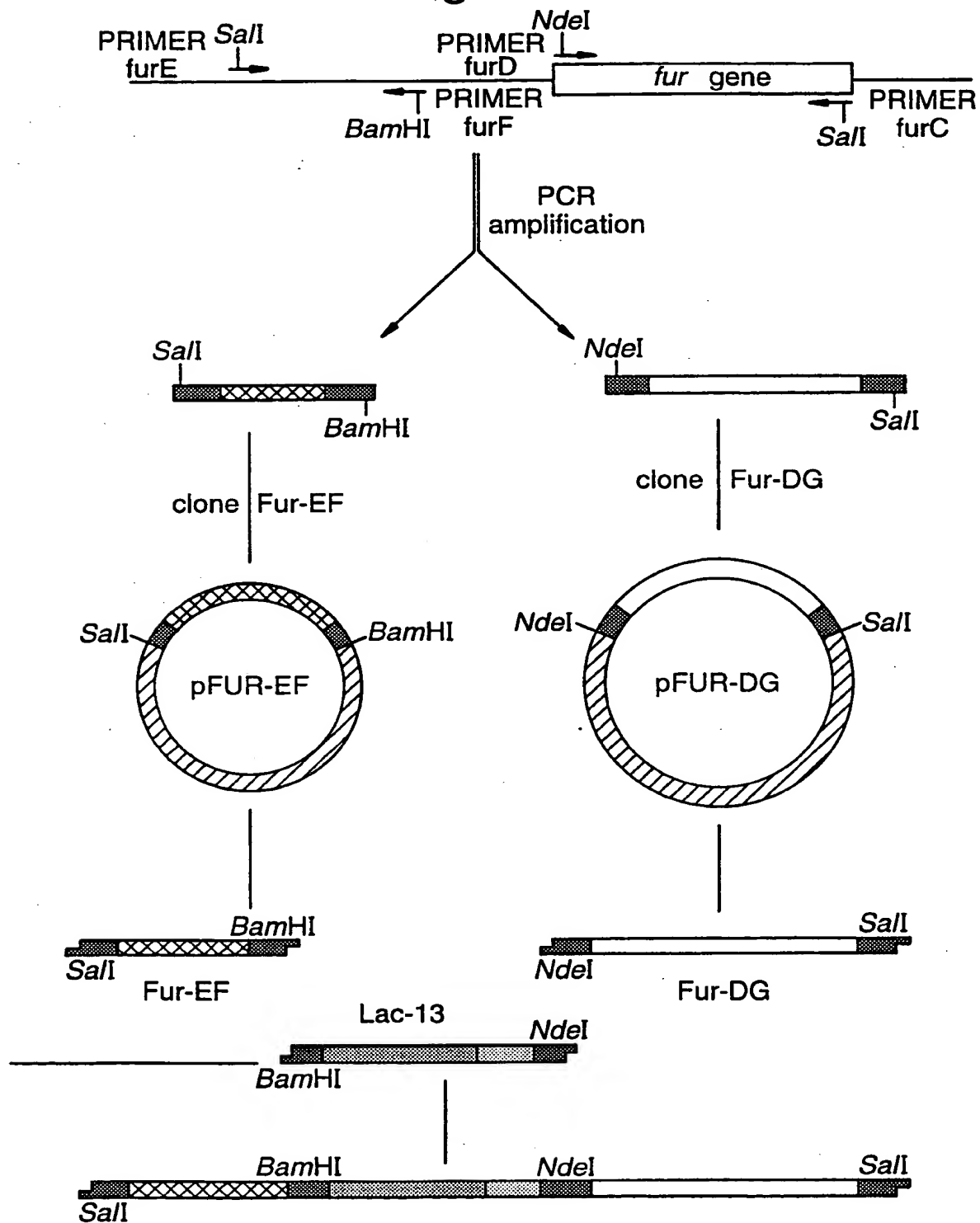
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Fig.5a.



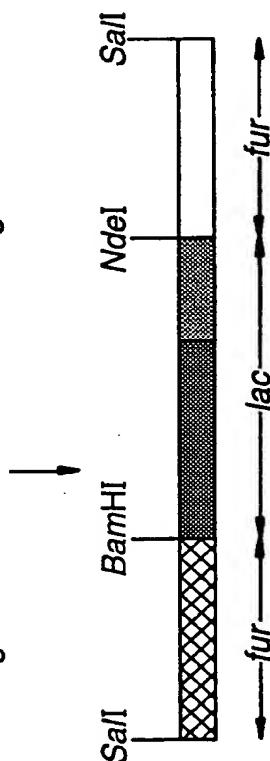
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Fig.5b.

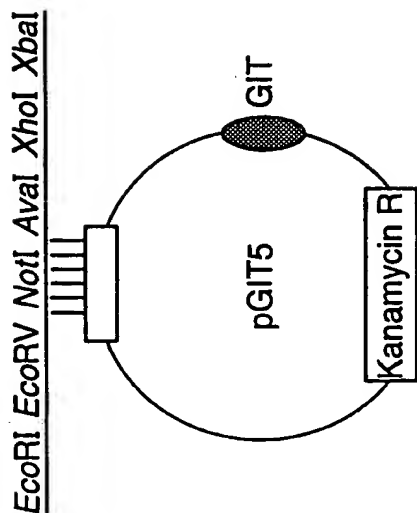


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The *fur/lac* construct was cut from the plasmid pUC18-*fur/lac* using *Sal*I. The 5' overhangs were filled in with klenow to give blunt ends.



The blunt ended *fur/lac* fragment was ligated into the blunt end *Eco*RV site of pGIT5 to give pGIT5*fur/lac*



pGIT5 was cut with the blunt end cutter *Eco*RV

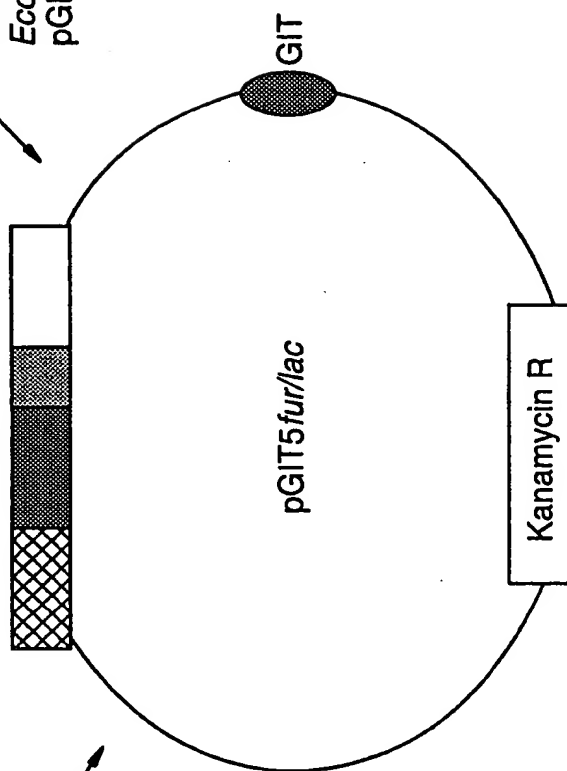


Fig.5c.

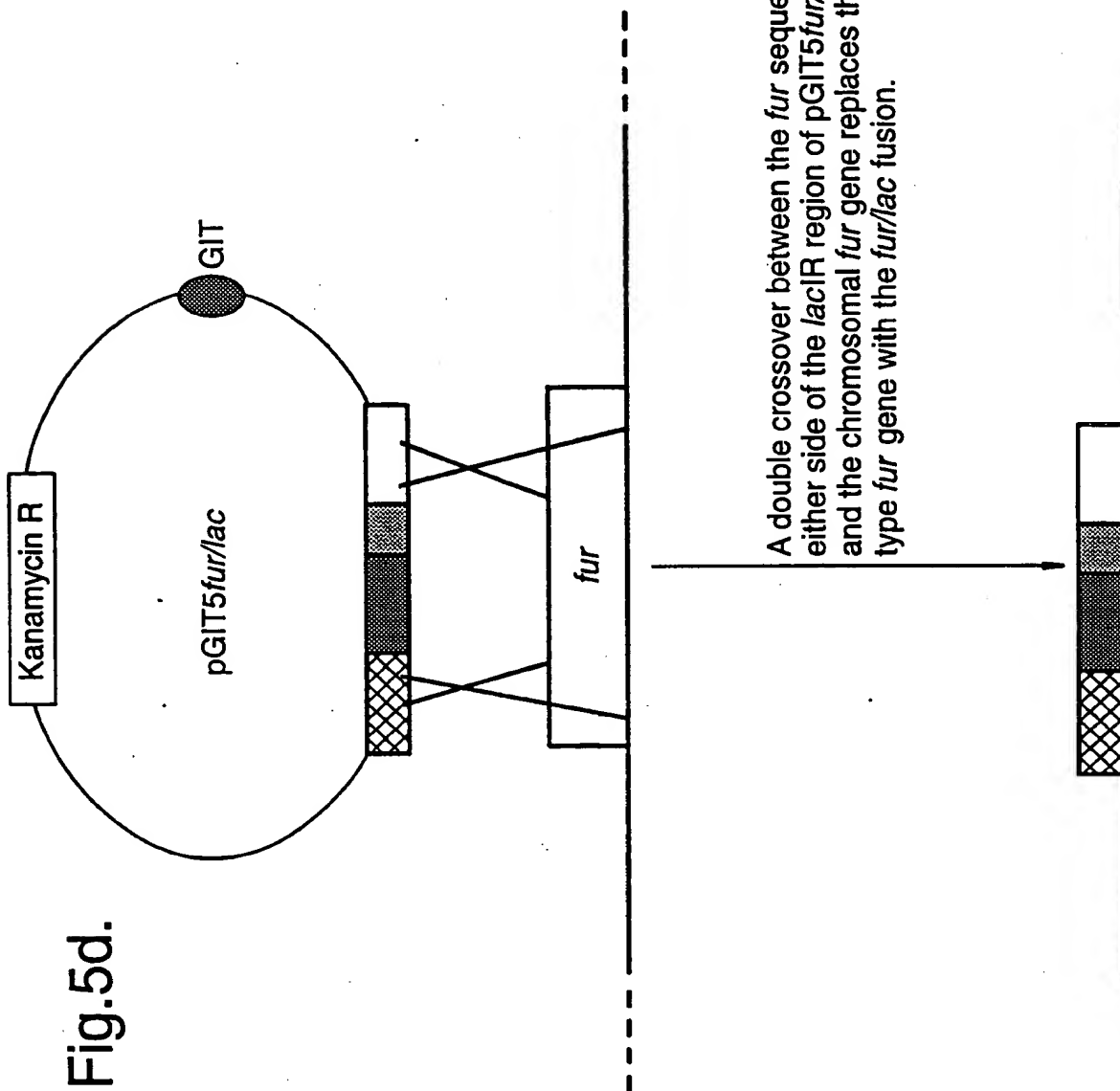


Fig.5d.